

FIGURE 1A

1 CGGCAGCAAAGGAACGTGCGAACGGTGACGCCGCCGACTGGCTCGGCTCTCCGTGC
61 CCCGGCGTCCTCCGCCGCTCATGGCCCGGCCGCGGACGAGCGGCGCTGAGGCGGG
121 CGCGTGGAGACGTGAGGCCGCCGCTGGCCCTCACAGTCGGCGTTTCGCCGCCCTGCC
181 GCGGTGCCGCCGCAACGCCATGCCCTCGCGCCTGGCTGGCGGGGCCGTGTCC
241 TCCCAGGCCGTCCGCCGCCCTGGAGCTCGGCCGCCGCCGCCGCCGCCGCCGCC
301 AGGCGCGAGGAGGCCGCCACCGCCGCCGCCGCCGCCGCCGCCGCCGCCGCCGCC
361 GTGGAGCAGAACGAGCTGCAGCAGAGCGGCCAGAAGCACCAGCAGACGTTTGAAAT
421 CAACTGAGGAAATTACGGGATAATGACACCCAGATACTACAGCAAGCCTGAAGGAT
481 AGTAATGAAACTTGAATTAGCAGTGGCTTCCTACTCGCAAGAAATGCTAAGACCCCT
541 CAGCAGGAGGAGACAACATTACCAAAACAGCACTTCCTGGCAATGATAGATACTCAGT
601 GTGGGAAGCCAAGCAGATAACAAATGTGATTGATCTCACTGGAGATGATAAAAGATGATCTT
661 CAGAGAACAAATTGCCCTGAGTTGGCCAACTCAAAACAGGGCATTAGGGAGACTGGAATA
721 ACTGATGAGGAACAAGCCATTAGCAGAGTTCTGAAAGCCAGTATAGCAGAGAATAAGCA
781 TGTTGAAGAGGACACCTACAGAAGTTGGAGGATTCTGAAACCCATTGATAGAAAAA
841 AGACAGGACAAAGCTCCCCTGGCTAAAGAATGTTGGCAATACTGTTGGTTAGTGCT
901 GTTATTCACTTATTAACTTTGGAATTAGAAGGATTAGTTCTGAATTACAAGCCT
961 CCATCAAATGCTCAAGATTACCCGAAACAAAAGGAACATCGGAATTGCCCTTTATG
1021 CGTGAGCTGAGGTATCTTGCACITCTTGTGTTGACAAAAGGAAGTATGTTGATCCA
1081 TCAAGAGCAGTTGAAATTCTTAAGGATGCTTCAAATCAAAATGACTCACAGCAGCAAGAT
1141 GTGAGTGAAGTTACACACAAATTATTAGATTGGTAGAAGATGCGCTTCAAATGAAAGCT
1201 GAAGAGGAGACGGATGAAGAGAACCCAAAGAACCCCATGGTAGAGTTCTATGGCAGA
1261 TTCCTGGCTGGGAGTACTTGAAGGTTAAAAAAATTGAAAACACTGAAATGTTGGTCAG
1321 TACCCACTTCAGGTCAATGGGTCAAAGATCTGCATGAGTGCCTAGAAGCTGCAATGATT
1381 GAAGGAGAAATTGAGTCTTACATTAGAATTCAGGAAATCAGGGCAAGAGCATTGG
1441 TTTACTGGATTACACCTGTGTTAACATTGANTTGTCAAGATTGAAATTAACTCAGGCA
1501 TTGGGAAGACCAAGAAAAATTCAACAAATTAGAATTTCACCAAGTTGAAACTTCCCAAGTTTATATTGGAC
1561 AGATACATGCACAGAAACAGAGAAATAACAAGAATTAGAGGGAAAGAGATCAAGAGACTG
1621 AAAGATTACCTCACGGTATTACAACAAAGGCTAGAAAGATATTAAAGCTATGGTCCGGT
1681 CCCAACGATTCCCCCTGGTAGATGTCAGTGCATTGGAAATTGCCCTCAAGTAAA
1741 CCTGTTGCACTTCTCTGTGACGATATTGACGCTAGTCCCCCACCTAGTGGTTCCATA
1801 CCATCACAGACATTACCAAGCACAAACAGAACACAGGGAGCCCTATCTCAGAACTGCCA
1861 AGCACATCACCTCATCAGTTGCTGCCATTTCATCGAGATCAGTAATACACAAACCAATT
1921 ACTCAGTCCGGATACCTCAGATTGCCCCATGCATCCGGCACCAAGGCACATAACGGAG
1981 GAAGAACATTCTGTGCTGGAAAGTTGTTACATCGCTGGAGGACAGAAATAGAAAATGAC
2041 ACCAGAGATTGCTGAGGAAAGCATATCCAGAAATCAGAACAAATTGAAATTAAATGACTCT
2101 GACAAATCTATGATACAAGTTCTTATCGATTACATGCCGTTTAGTTCACTGAAAGGCCAA
2161 GCTAATGCTGGCACTACTGGCATATATTGATCATCGTAAAGCAGATGGATGAAG
2221 TACAATGATATTGCTGTGACAAAATCATCATGGAAAGAGCTAGTGAGGGACTTTGGT
2281 GGTATAGAAATGCCAGTCAGACTGTTAATGACATAAAATGATAAGGCACAGTTCTA
2341 ATACAAGAGGAGTTAATAAGAAACTGGCAGCCCCCTGGTAGAAGAAACATTACCA
2401 CCGGATTGAGAGATTGTTGAGGAAGACAACCAACGATTGAAAAGAAACTAGAAGAA
2461 TGGGATGCACAACCTTGCCTGGAAAGCTTGCAGGAAAAGCTTTAGCGTCTCAGAAATTG
2521 AGAGAGTCAGAGACTTCTGTGACAACAGCACAAGCAGCAGGAGACCCAGAATATCTAGAG
2581 CAGCCATCAAGAAGTGAATTCTCAAGCACTTGAAGAAGAAACTATTCAAATAATTACC
2641 AAGGCATCACATGAGCATGAAGATAAAAGCTGTAACAGTTGAGTCAGTCGGCAATTAG
2701 TTGGAATATGCAAGGTTGGTTAAGTGGCCAAGAAGACACCCACAGAAACCGATTAT
2761 CGTTTACATCATGTTAGTGGCTACTTTATCCAGAACCCAGGGACCAAGAAAATTATTGAG
2821 AAAACATTACTAGAACAAATTGGAGATAGAAATTGAGTTGATGAAAGGTGTACAAC
2881 ATAATGAAAGTTGCTCAAGCCAAACTGGAAATGATAAAACCTGAAGAAGTAAACTGGAG
2941 GAATATGAGGAGTGGCATCAGGATTATAGGAAATTCAAGGAAACAACATATGTATCTCATA
3001 ATTGGGCTAGAAAATTTCACAAAGAGAAAGTTATATAGATTCTTGTGTTCTCATCTGT
3061 GCTTATCAGAATAACAAAGAACTCTTGTCTAAAGGCTTATACAGAGGACATGATGAAGAA
3121 TTGATATCACATTATAGAAGAGAAATGTTGCTAAATTAAATGAGCAAGCCGAGAACTC
3181 TTCGAATCTGGAGAGGATCGAGAAGTAAACAATGGTTGATTATCATGAATGAGTTATT
3241 GTCCCATTTTGCCTTATTACTGGTGGATGAAATGGAAGAAAAGGATATACTAGCTGTA
3301 GAAGATATGAGAAATGATGGTGTCTACCTGGTAGAAGAAATGGAACCACACCTCCAA
3361 GAAAAGCTGACAGATTGGCCAAAATGCTGATTGTTCTATGGAGATTAAAGTTTC

3421 CATGAGCCACCGAAGTTACCTTCATATTCCACGCATGAACACTGTGAGCGATTGCCGA
3481 ATCATGTTGCCCTCAGTCGAACCTCCTGCTGATGGAAGATAAAACTGCACACTTCCCTGA
3541 ACACACTGTATAAACCTTTAGTTCTAACCCCTGCCTCCTGTCACAGGGTTGCTT
3601 GTTGCTGCTATAGTTAACCTTTTATTAAATAACTGCAAAAGACAAAATGACTA
3661 TACAGACTTTAGTCAGACTGCAGACAATAAAGCTGAAAATCCGATGGCGCTCAGACATTT
3721 TAACCGGAACTGATGTATAATCACAAATCTAATTGATTTATTATGGCAAAACTATGCTT
3781 TTGCCACCTCCTGTTGCAGTATTACTTGCTTTATCTTTCTTCTAACAGCTTCC
3841 ATTCAAGTCTGGATCCTCCATGACTACAGCCATTAAAGTGTTCAGCACTGTGTACGATAC
3901 ATAATATTGGTAGCTTGTAAATGAAATAAAGTTTATTATGGCTAC

FIGURE 1B

FIGURE 2

1 MTVEQNVLQQSAAQKHQQTFLNQLREITGINDTQILQQALKDSNGNLELAVAFLTAKNAK
61 TPQQEETYYQTALPGNDRYISVGSQADTNVIDLTGDDKDDLQRTIALSLAESNRAFRET
121 GITDEEQAIISRVLEASIAENKACLKRTPTEVWRDSRNPYDRKRQDKAPVGLKNVGNTCWF
181 SAVIQSLFNLLEFRRLVLYKPPSNAQDLPRNQKEHRNLPFMRELRYLFALLVGTKRKYV
241 DPSRAVEILKDAFKSNDSQQQDVSEFTHKLLDWLEAFQMKAEETDEEKPKPMVELFY
301 GRFLAVGVLEGKKFENTEMFGQYPLQVNGFKDLHECLEAAMIEGEIESLHSENSGKSGQE
361 HWFTGLPPVLTFXLSRFEFNQALGRPEKIHNKLEFPQVLYLDRYMRNREITRIKREEIK
421 RLKDYLTVLQQRLERYLSYSGSPKRFPLVQYALEFASSKPVCSPVDDIDASSPPSG
481 SIPSQTLPSSTTEQQGALSSelpstSPSSVAISSLRSVIHKPFTQSRIPPDLPMPHPAPRHI
541 TEEELSVLESCLHRWRTEIENDTRDLQESISRIHRTIELMYSKSMIQVPYRLHAVLVHE
601 GQANAGHYWAYIFDHRESRWMKYNDIAVTKSSWEELVRDSFGGYRNASAYCLMYINDKAQ
661 FLIQEEFNKETGQPLVGIETLPPDLRDVEEDNQRFEKELEEWDAQLAQKALQEKL LASQ
721 KLRESETSVTTAQAAAGDPEYLEQPSRSDFSKHLKEETIQIITKASHEHEDKSPETVLQSA
781 IKLEYARLVKLAQEDTPPETDYRLHHVVYFIQNQAPKKIIETKLLEQFGDRNLSFDERC
841 HNIMKVAQAKLEMICKPEEVNLEEEWHDYRKFRRETTMYLIIGLENFQRESYIDSLLFL
901 ICAYQNNKELLSKGLYRGHDEELISHYRRECLLKLINEQAAELFESGEDREVNNGLIIMNE
961 FIVPFLPLLLVDEMEEKDILAVEDMRNRWCSYLGQEMEPHLQEKLTDFLPKLLDCSMEIK
1021 SFHEPPKLPSSYTHELCERFARIMLSRTPADGR

FIGURE 3A

1 CGGCAGCAAAGAACGTGCGAACCGCGTACGCCGCCGACTGGCTCGCCTCTCCCGTGC
61 CCCGGCGTCCTCCGCCGCTATGGCCGGGCCGCGGACAGCGCGCTGAGGCGGG
121 CCGCGTGGAGACGTGAGGCCGCCGCGCTGGCCCTCACAGTCGGCTTCGCCGCCCTGCC
181 GCGGTGCCCGCGCACGCCATGCCCTCGCCTGGCTGGCGGGCGCTGTCC
241 TCCCAGGCCGTCCGCCGCCCTGGAGCTGGCGAGCGCGCAGCCAGGCCGG
301 AGGCAGGAGGCCGCCGCCACCGCCGCCGCCGCCGCCGCCGCCGCCATGACC
361 GTGGAGCAGAACGTGCTGCAGCAGAGCGCGCAGAAGCACCAGCAGACGTTTGAAAT
421 CAACTGAGAGAAATTACGGGATTAAATGACACCCAGATACTACAGCAAGCCTTAAGGAT
481 AGTAATGAAACTTGAATTAGCAGTGGCTTCCTTAAGCAGAAGAATGTAAGACCCCT
541 CAGCAGGAGGAGACAACCTACTACCAAACAGCACTCCTGGCAATGATAGATAACATCAGT
601 GTGGGAAGCCAAGCAGATAACAAATGTGATTGATCTCACTGGAGATGATAAGATGATCTT
661 CAGAGAACAAATTGCCCTGAGTTGGCGAATCAAACAGGGATTCAAGGGAGACTGGAATA
721 ACTGATGAGGAACAAGCATTAGCAGAGTTCTGAAGCCAGTATAGCAGAGAATAAGCA
781 TGTGAGGAGGACACCTACAGAACGTTGGAGGGATTCTCGAAACCCATTATGATAGAAAA
841 AGACAGGACAAAGCTCCGTTGGCTAAAGAATGTTGCAATACTTGTGTTAGTGCT
901 GTTATTCACTCATTTAACACACCCGAAACCAAAAGGAACATCGGAATTGCCCTTATG
961 CCATCAAATGCTCAAGATTACCCGAAACCAAAAGGAACATCGGAATTGCCCTTATG
1021 CGTGAGCTGAGGTATCTATTGCACTTCTGTTGACCAAAAGGAAGTATGTTGATCCA
1081 TCAAGAGCAGTGAATTCTTAAGGATGCTTCAAAATGACTCACAGCAGCAAGAT
1141 GTGAGTGAAGTTACACACAAATTATTAGATTGGTTAGAAGATGCCCTCAAATGAAAGCT
1201 GAAGAGGAGACGGATGAAGAGAAGCAGAACCCCCATGGTAGAGTTGTTATGGCAGA
1261 TTCCCTGGCTGTGGGAGTACTTGAAGGTAAAAATTGAAAACACTGAAATGTTGGTCAG
1321 TACCCACTTCAGGTCAATGGTTCAAAGATCTGCATGAGTGCCTAGAAGCTGCAATGATT
1381 GAAGGAGAAATTGAGTCTTACATTCAAGAAATTCAAGGAAATCAGGCAAGAGCATTGG
1441 TTTACTGGATTACCACCTGTGTTAACATTGANTTGTCAAGATTGAAATTAAATCAGGCA
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1681 CCCAAACGATTCCCCCTGGTAGATGTTCTCAGTATGCATTGAAATTGCCCTCAAGTAA
1741 CCTGTTGCACTCTCCTGTTGACGATATTGACGCTAGTCCCCACCTAGTGGTCCATA
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1861 AGCACATCACCTCATCAGTGTGCCATTCACTCGAGATCAGTAATACACAAACCAATT
1921 ACTCAGTCCGGATAACCTCAGATTGCCCCATGCATCCGGCACCAAGGCACATAACGGAG
1981 GAAGAACATTCTGTGCTGGAAAGTTGTTACATCGCTGGAGGACAGAAATAGAAATGAC
2041 ACCAGAGATTGCAAGGAAAGCATACTCCAGAATCCATCGAACAAATTGAATTATGTA
2101 GACAATCTATGATAACAAGTCCCTATCGATTACATGCCGTTTTAGTTCACGAAGGCCA
2161 GCTAATGCTGGGCACTACTGGGATATATTGATCATCGAAAGCAGATGGATGAAG
2221 TACAATGATATTGCTGTGACAAAATCATCATGGAAAGAGCTAGTGAGGGACTCTTGGT
2281 GGTTATAGAAATGCCAGTGCATACTGTTAATGACATAAAATGATAAGGCACAGTCC
2341 ATACAAGAGGAGTTAATAAAGAAACTGGGAGGCCCTGTTGAGTAAAGACATTACCA
2401 CCGGATTTGAGAGATTGTTGAGGAAGACAACCAACGATTGAAAAGAAACTAGAAGAA
2461 TGGGATGCACAACCTGCCAGAAAGCTTGCAGGAAAAGCTTTAGCGTCTCAGAAATTG
2521 AGAGAGTCAGAGACTTCTGTGACAACAGCACAAGCAGCAGGAGACCCAGAATATCTAGAG
2581 CAGCCATCAAGAAGTGATTCTCAAAGCACTTGAAGAGAAAGAAACTATTCAAATAATTAC
2641 AAGGCATCACATGAGCATGAAGATAAAAGTCTGAAACAGATTGTTGCAGTCGGCAATTAAG
2701 TTGGAATATGCAAGGTTGTTAAGTTGCCAAGAAGACACCCACCAAGAAACCGATTAT
2761 CGTTACATCATGAGTGGCTACTTATCCAGAACCAAGGCCACAAAGAAAATTATTGAG
2821 AAAACATTACTAGAACATTGGAGATAGAAATTGAGTTGATGAAAGGTGTACAAC
2881 ATAATGAAAGTTGCTCAAGCAAACCTGGAAATGATAAAACCTGAAGAAGTAAACTTGGAG
2941 GAATATGAGGAGTGGCATCAGGATTAGGAAATTAGGGAAACAAACTATGTATCTCATA
3001 ATTGGGCTAGAAAATTTCAAAGAGAAAGTTATAGATTCCCTGCTGTTCCATCTGT
3061 GCTTATCAGAATAACAAAGAACTCTGTCTAAAGGTTATACAGAGGACATGATGAAGAA
3121 TTGATATCACATTAGAACAGGATGTTGCTAATCCTTAATTAAAAGGAACAAAAAC
CTATTCTTTTTCTGCATTGCAAGAAATTAAATGAGCAAGCCGCAGAACTC
3181 TTCGAATCTGGAGAGGATCGAGAAGTAAACAATGGTTGATTATCATGAATGAGTTATT
3241 GTCCCATTGGCATTATTACTGGTGGATGAAATGGAAGAAAAGGATATACTAGCTGTA
3301 GAAGATATGAGAAATCGATGGTGTCTACCTGGTCAAGAAATGGAACCAACACCTCAA
3361 GAAAAGCTGACAGATTGGCCAAAATGCTGATTGAGATTAAAAGTTTC

3421 CATGAGCCACCGAAGTTACCTCATATTCCACGCATGAACCTCTGTGAGCGATTGCCCGA
3481 ATCATGGTGCCTCAGTCGAACCTCTGCTATGGAAGAACTGCACACTTCCCTGA
3541 ACACACTGTATAAACTCTTTAGTTCTAACCCCTGCCTCTGTACAGGGTTGCTT
3601 GTTGCTGCTATAGTTTAACCTTTTATTTAATAACTGAAAAGACAAAATGACTA
3661 TACAGACTTAGTCAGACTGCAGACAATAAGCTGAAAATCGCATGGCGCTCAGACATT
3721 TAACCGGAACTGATGTATAATCACAATCTAATTGATTTATTATGGCAAAACTATGCTT
3781 TTGCCACCTCCTGTTGCAGTATTACTTTGCTTTATCTTCTCAACAGCTTCC
3841 ATTCACTCTGGATCCTCCATGACTACAGCCATTAAAGTGGTACGACTGTGTACGATAC
3901 ATAATATGGTAGCTTGTAAATGAAATAAAGTTTATTTATGGCTAC

FIGURE 3B

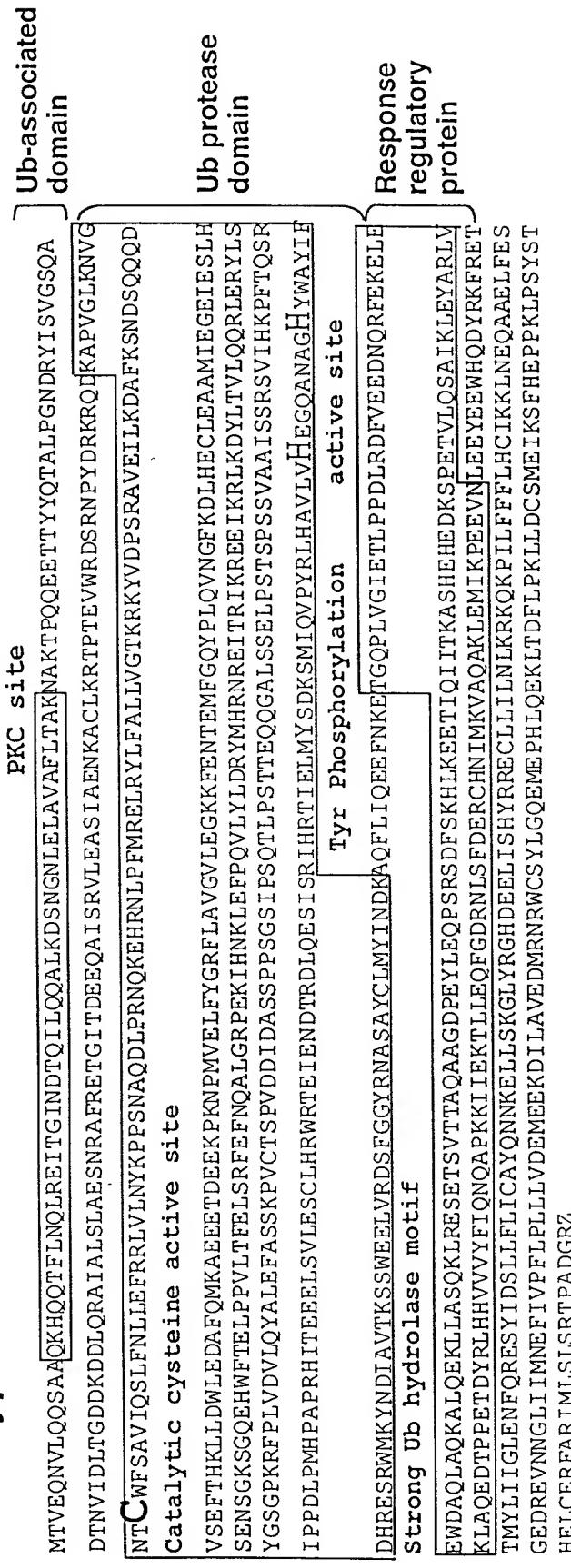
FIGURE 4

1 MTVEQNVLQQSAAQKHQQTFLNQLREITGINDTQILQQALKDSGNLELAVAFLTAKNAK
61 TPQQEETYYQTALPGNDRYISVGSQADTNVIDLTGDDKDDLQRTIALSLAESNRARET
121 GITDEEQAIISRVLEASIAENKACLKRTPTEVWRDSRNPYDRKRQDKAPVGLKNVGNTCW
181 SAVIQSLFNLLEFRRRLVLNYKPPSNAQDLPNQKEHRNLPFMRELRYLFALLVGTKRKYV
241 DPSRAVEILKDAFKSNDSSQQDVSEFTHKLLDWLEDAFQMKAEEETDEEKPKNPMVELFY
301 GRFLAVGVLEGKKFENTEMFGQYPLQVNNGFKDLHECLEAAMIEGEIESLHSENSGKGQE
361 HWFTGLPPVLTFXLSRFEFNQALGRPEKIHNKLEFPQVLYLDRYMHNRNREITRIKREEIK
421 RLKDYLTVLQQRLERYLSYGGSPKRFPLVDVLQYALEFASSKPVCSTPVDDIDASSPPSG
481 SIPSQTLPSSTTEQQGALSSELPSTSPSSVAAISSRSVIHKPFTQSRIPPDLPMPAPRHI
541 TEEELSVLESCLHWRTEIENDTRDLQESISRHIERTIELMYSDKSMIQVPYRLHAVLVHE
601 GQANAGHYWAYIFDHRESRWMKYNDIAVTKSSWEELVRDSFGGYRNASAYCLMYINDKAQ
661 FLIQEEFNKETGQPLVGIETLPPDLRDVEEDNQRFEKELEEWDAQLAQKALQEKL LASQ
721 KLRESETSVTTAQAAAGDPYEYLEQPSRSDFSKEETIQIITKASHEHEDKSPETVLSQA
781 IKLEYARLVKLAQEDTPPETDYRLHHVVVFIQNQAPKKIEKTLLEQFGDRNLSFDERC
841 HNIMKVAQAKLEMICKPEEVNLEEEWHQDYRKFRETTMYLIIGLENFQRESYIDSLLFL
901 ICAYQNNKELLSKGLYRGHDEELISHYRRECLLILNLKRKQKPILFFFHLHCIKKLNEQAA
961 ELFESGEDREVNNGLIIMNEFIVPFLPLLVDEMEEKDILAVEDMRNRWCSYLGQEMEPH
1021 LQEKLTDFLPKLDCSMEIKSFHEPPKLPYSTHELcerfarimlslsrtPADGR

Figure 5

Sequence of SUP

Wild type SUP



Mutant SUP

Cys → Ser

mtSUP Suppresses α -IgM-Induced NFAT-Luciferase Activity as a Dominant-negative Mutant

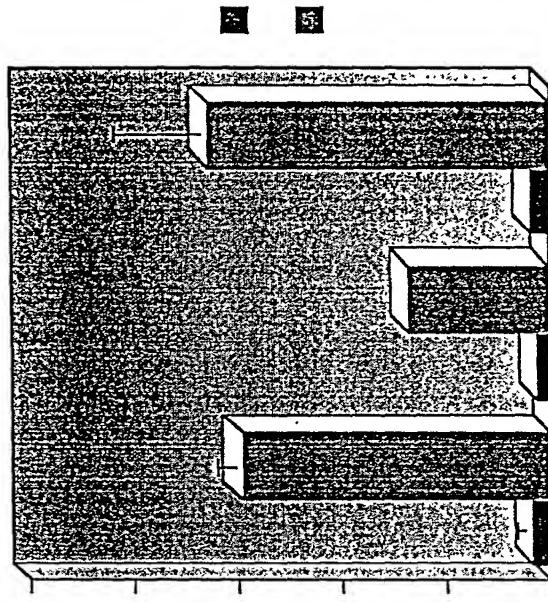
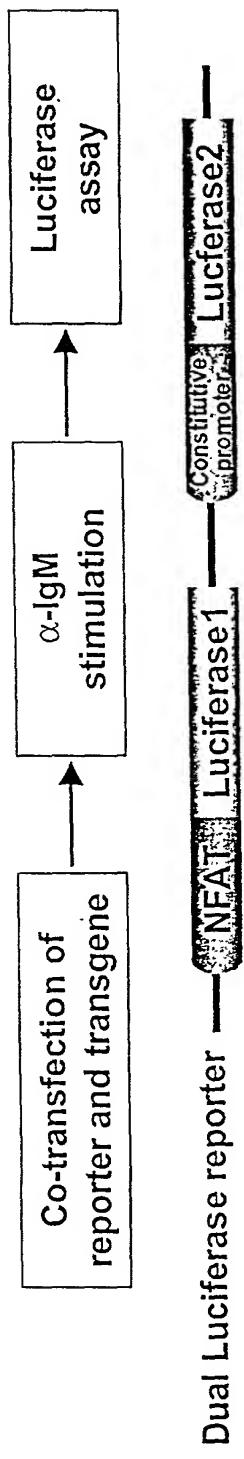
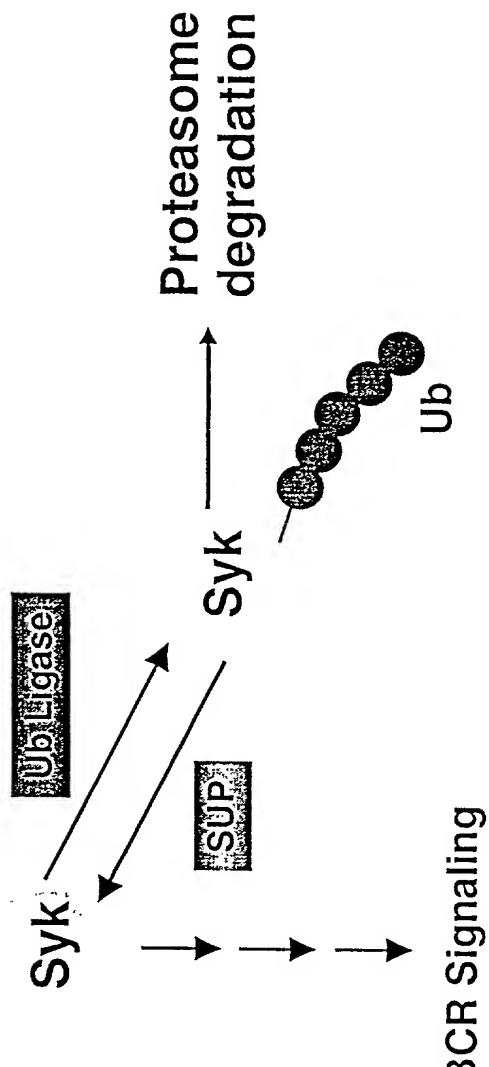


Figure 7

Model: SUP Regulates BCR Signaling by Stabilizing Syk



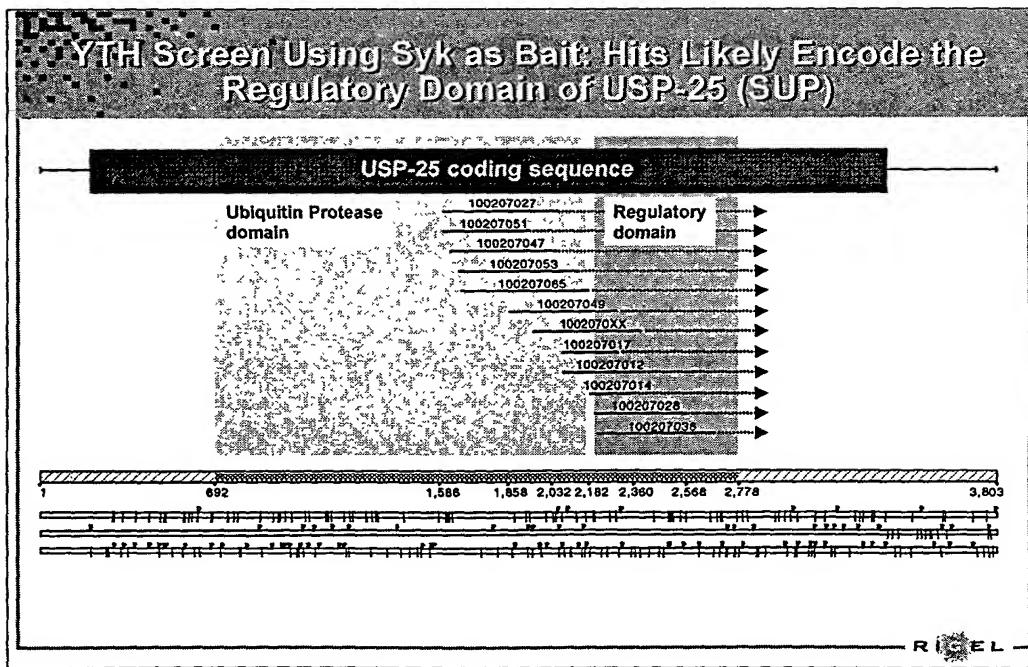
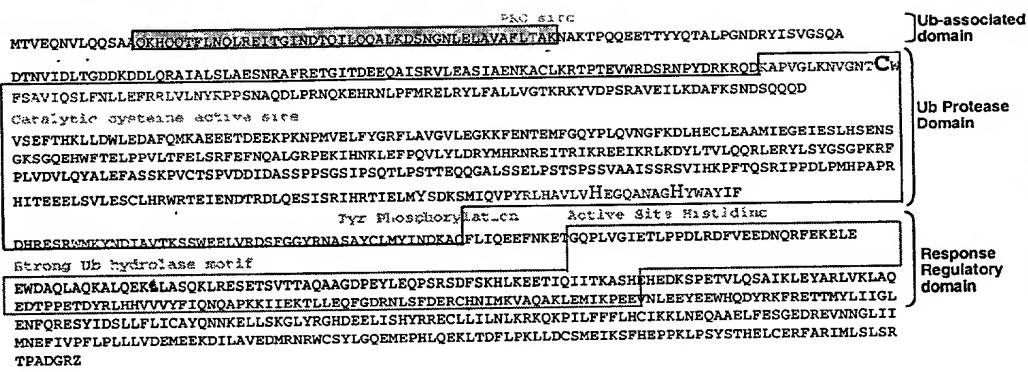


Fig. 8

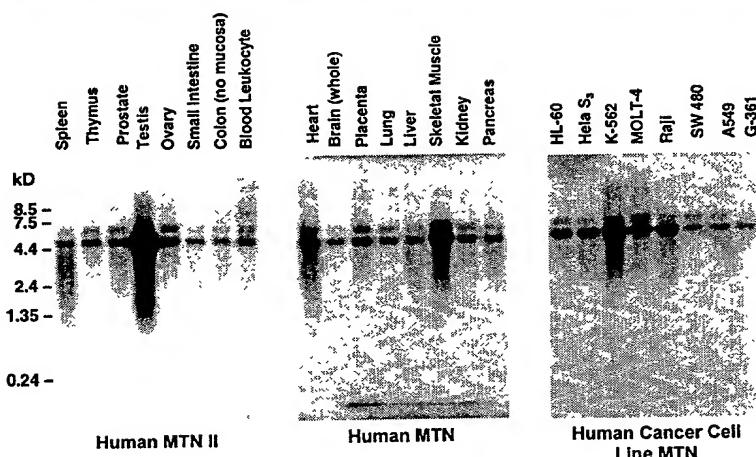
Syk Interacting Ubiquitin Protease USP-25

Wild type USP-25



RIGEL

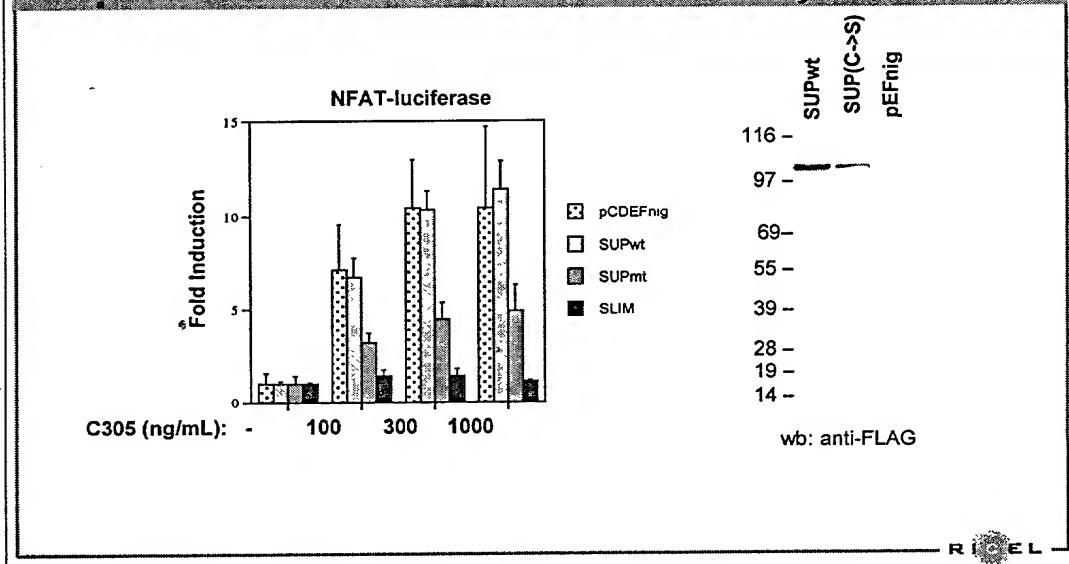
USP-25 mRNA Expression Profile



RIGEL

Fig. 9

Catalytically Inactive USP-25(C->S) blocks TCR-induced NFAT activity



Catalytically Inactive USP-25 Inhibits Downstream TCR Signaling

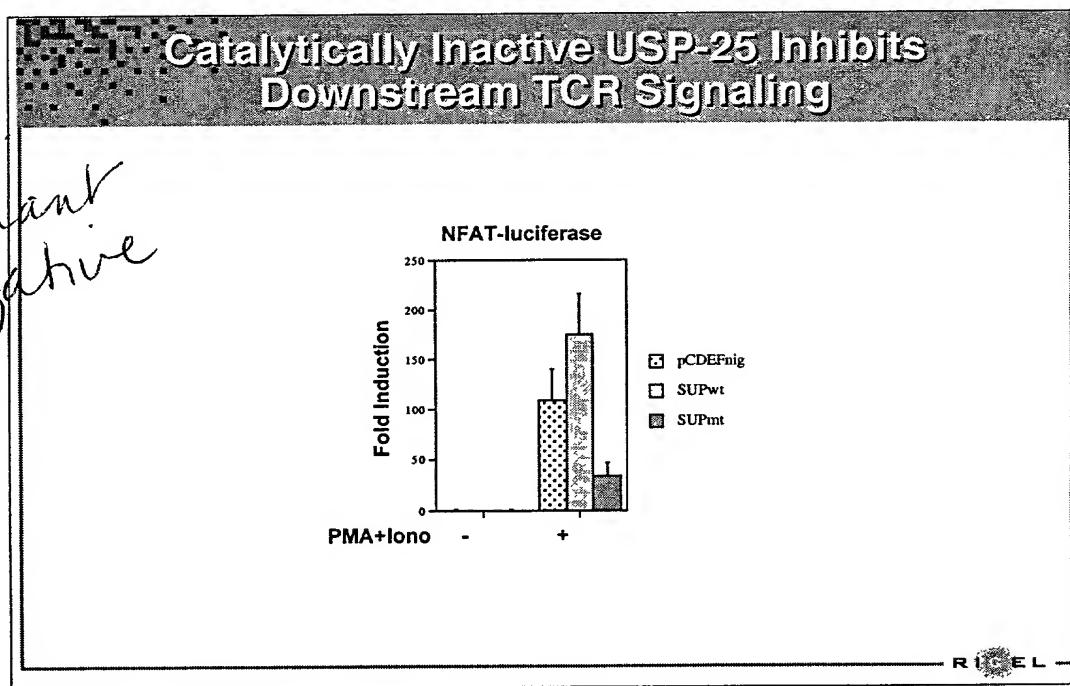


Fig. 10

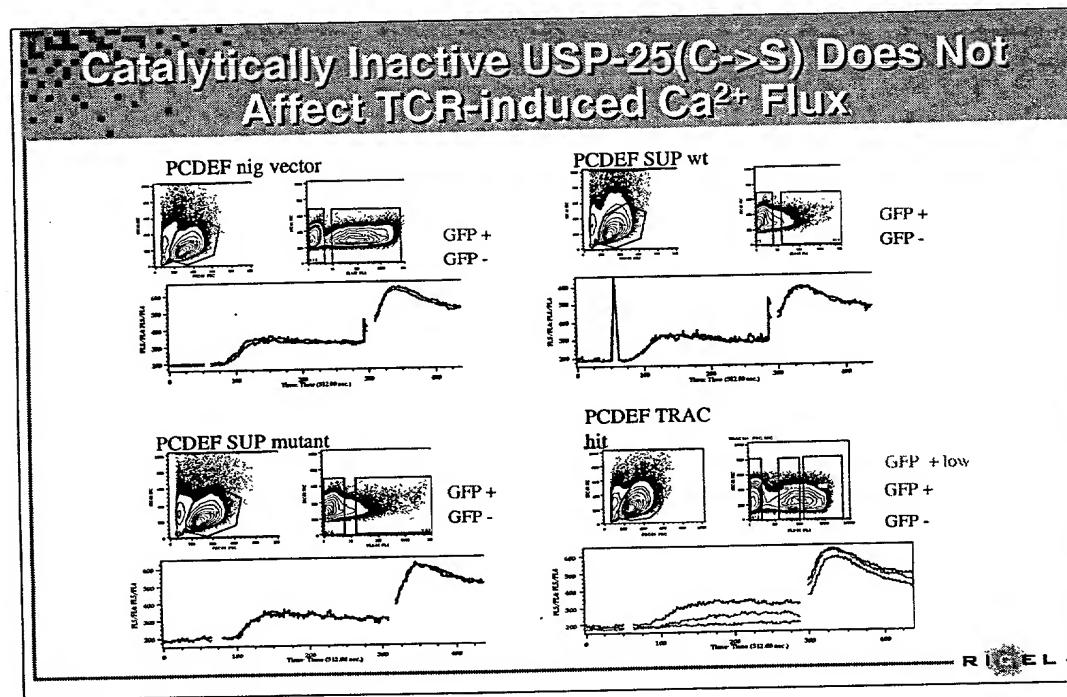
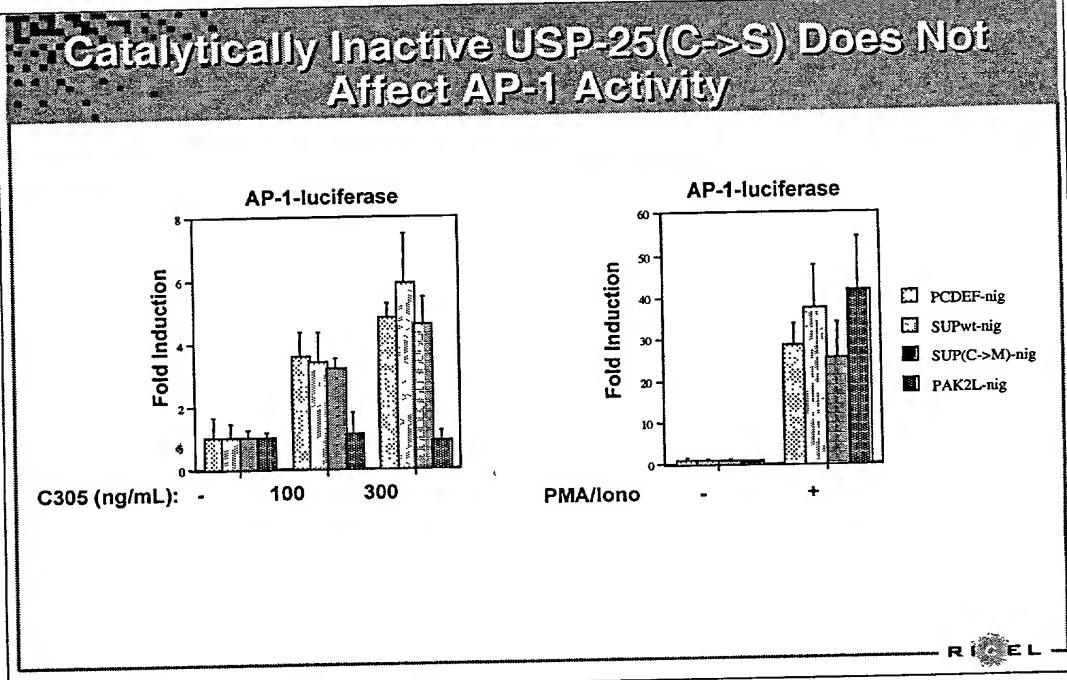


Fig. 11

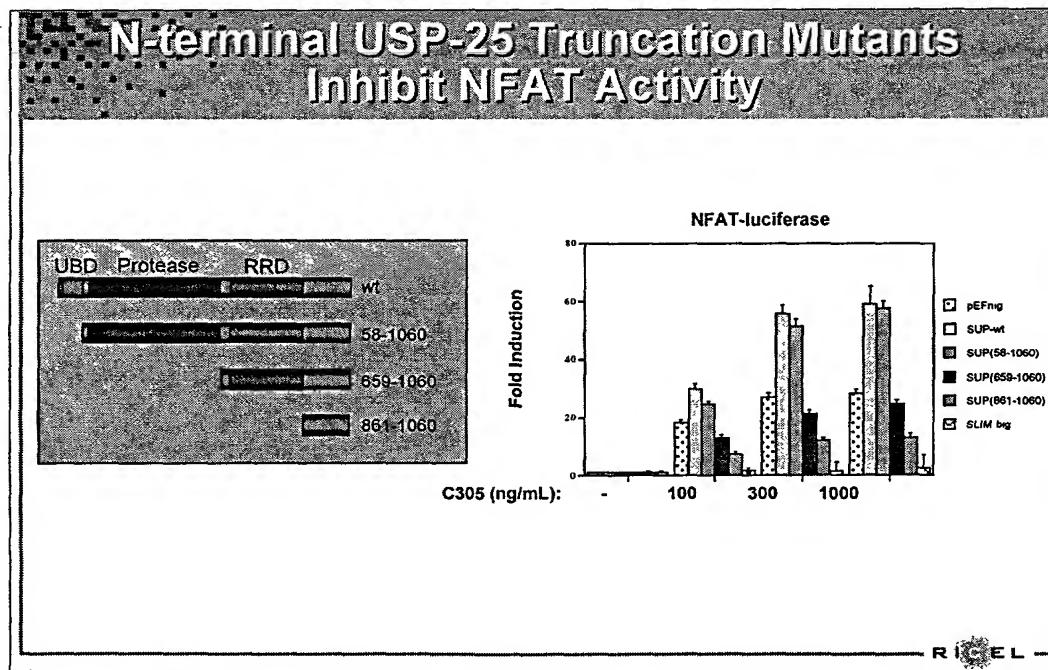
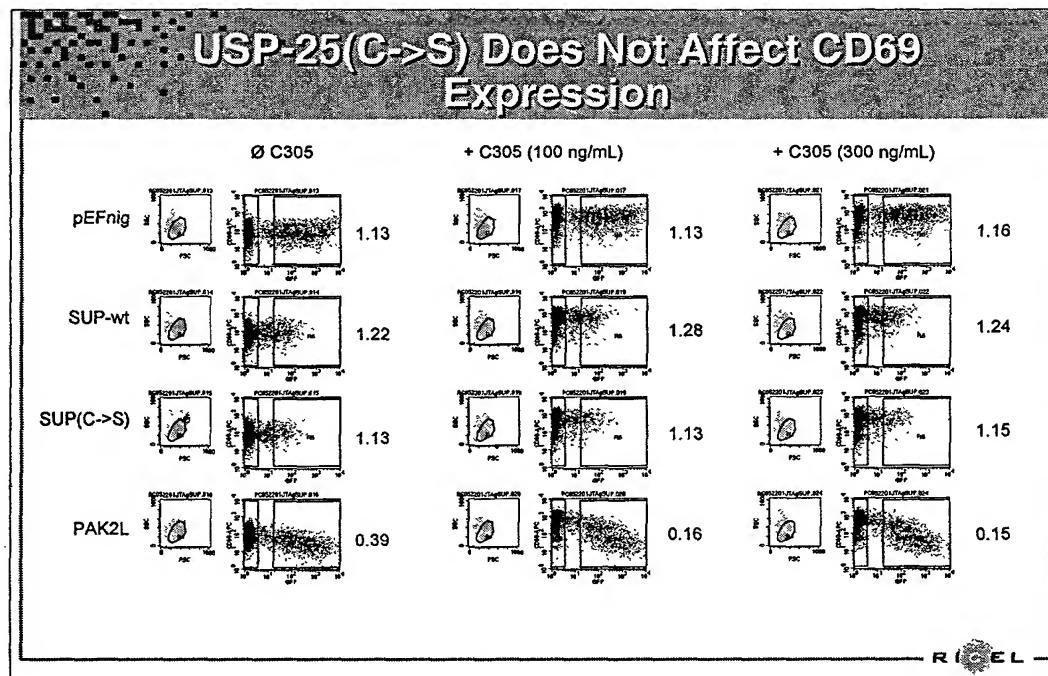
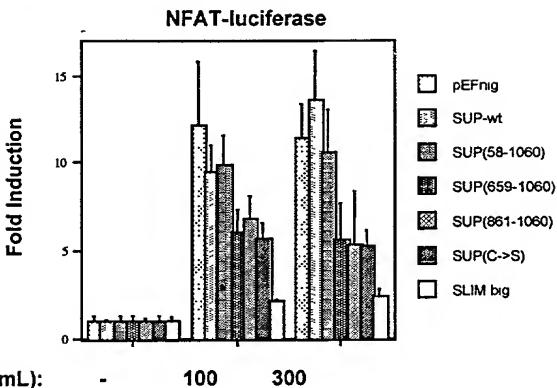


Fig. 12

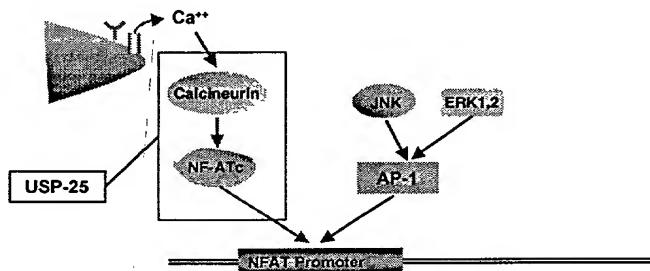
USP-25 Mutants Inhibit NFAT activity in BJABs



R I C E L

Possible Regulatory Role of USP-25 on NFAT Activity

- USP-25 likely regulates the NFAT promoter downstream of Ca^{2+} and independent of the AP-1 pathway



R I C E L

Fig. 13

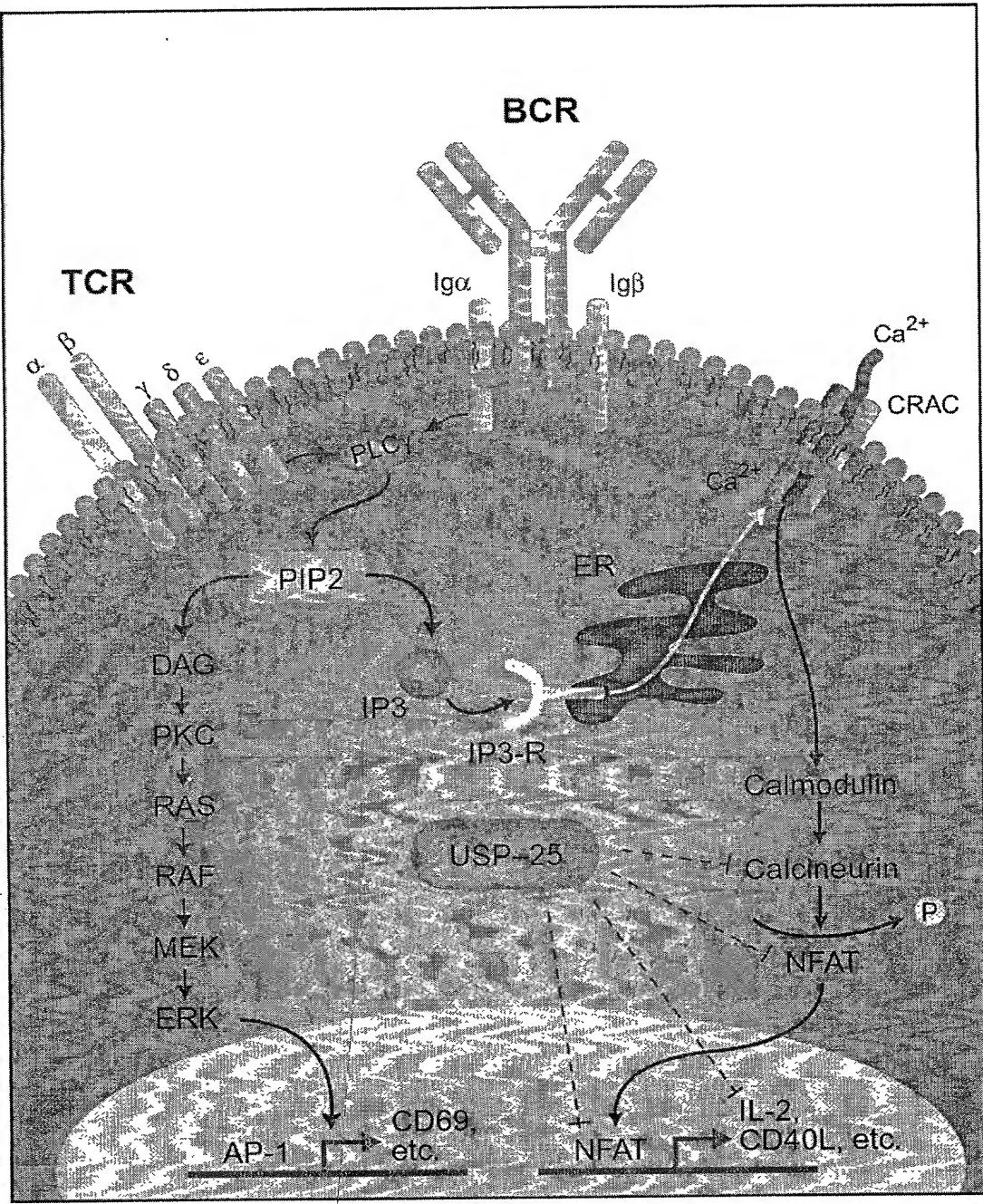


Fig. 14